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☐ 1: Protein Sci. 2007 May;16(5):990-5.

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**Structural context for protein N-glycosylation in bacteria:
The structure of PEB3, an adhesin from Campylobacter jejuni.**

**Rangarajan ES, Bhatia S, Watson DC, Munger C,
Cygler M, Matte A, Young NM.**

Campylobacter jejuni is unusual among bacteria in possessing a eukaryotic-like system for N-linked protein glycosylation at Asn residues in sequons of the type Asp/Glu-Xaa-Asn-Xaa-Ser/Thr. However, little is known about the structural context of the glycosylated sequons, limiting the design of novel recombinant glycoproteins. To obtain more information on sequon structure, we have determined the crystal structure of the PEB3 (Cj0289c) dimer. PEB3 has the class II periplasmic-binding protein fold, with each monomer having two domains with a ligand-binding site containing citrate located between them, and overall resembles molybdate- and sulfate-binding proteins. The sequon around Asn90 is located within a surface-exposed loop joining two structural elements. The three key residues are well exposed on the surface; hence, they may be accessible to the PglB oligosaccharyltransferase in the folded state.

PMID: 17456748 [PubMed - in process]

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Regulation of N-linked core glycosylation: use of a site-directed mutagenesis approach to identify Asn-Xaa-Ser/Thr sequons that are poor oligosaccharide acceptors. [Biochem J. 1997]

The amino acid following an asn-X-Ser/Thr sequon is an important determinant of N-linked core glycosylation efficiency. [Biochemistry. 1998]

N-linked glycosylation of rabies virus glycoprotein. Individual sequons differ in their glycosylation efficiencies and influence on cell surface expression. [J Biol Chem. 1992]

Identification of N-acetylgalactosamine-containing glycoproteins PEB3 and CgpA in Campylobacter jejuni. [J Clin Microbiol. 2002]

The hydroxy amino acid in an Asn-X-Ser/Thr sequon can influence N linked core glycosylation efficiency and the level of expression of a cell surface glycoprotein. [J Biol Chem. 1995]

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Multiple N-acetyl neuraminic acid synthetase (neuB) genes in *Campylobacter jejuni*: identification and characterization of the gene involved in sialylation of lipo-oligosaccharide.

Linton D, Karlyshev AV, Hitchen PG, Morris HR, Dell A, Gregson NA, Wren BW.

Department of Neurology, United Medical and Dental School, Guy's Hospital, London SE1 9RT, UK.

N-acetyl neuraminic acid (NANA) is a common constituent of *Campylobacter jejuni* lipo-oligosaccharide (LOS). Such structures often mimic human gangliosides and are thought to be involved in the triggering of Guillain-Barre syndrome (GBS) and Miller-Fisher syndrome (MFS) following *C. jejuni* infection. Analysis of the *C. jejuni* NCTC 11168 genome sequence identified three putative NANA synthetase genes termed neuB1, neuB2 and neuB3. The NANA synthetase activity of all three *C. jejuni* neuB gene products was confirmed by complementation experiments in an *Escherichia coli* neuB-deficient strain. Isogenic mutants were created in all three neuB genes, and for one such mutant (neuB1) LOS was shown to have increased mobility. *C. jejuni* NCTC 11168 wild-type LOS bound cholera toxin, indicating the presence of NANA in a LOS structure mimicking the ganglioside GM1. This property was lost in the neuB1 mutant. Gas chromatography-mass spectrometry and fast atom bombardment-mass spectrometry analysis of LOS from wild-type and the neuB1 mutant strain demonstrated the lack of NANA in the latter. Expression of the neuB1 gene in *E. coli* confirmed that NeuB1 was capable of in vitro NANA biosynthesis through condensation of N-acetyl-D-mannosamine and phosphoenolpyruvate. Southern analysis demonstrated that the neuB1 gene was confined to strains of *C. jejuni* with LOS containing a single NANA residue. Mutagenesis of neuB2 and neuB3 did not affect LOS, but neuB3 mutants were aflagellate and non-motile. No phenotype was evident for neuB2 mutants in strain NCTC 11168, but for strain G1 the flagellin protein from the neuB2 mutant showed an apparent reduction in molecular size relative to the wild type. Thus, the neuB genes of *C. jejuni* appear to be involved in the biosynthesis of at least two distinct surface structures: LOS and flagella.

PMID: 10712693 [PubMed - indexed for MEDLINE]

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Phase variation of a beta-1,3 galactosyltransferase involved in generation of the ganglioside GM1-like lipo-oligosaccharide of *Campylobacter jejuni*. [Mol Microbiol. 2000]

The sialic acid residue is a crucial component of *C. jejuni* lipooligosaccharide ganglioside mimicry in the induction of Guillain-Barre syndrome. [Neuroimmunol. 2006]

Structural characterization of *Campylobacter jejuni* lipooligosaccharide outer cores associated with Guillain-Barre and Miller Fisher syndromes. [Infect Immun. 2007]

Sialylation of lipooligosaccharide cores affects immunogenicity and serum resistance of *Campylobacter jejuni*. [Infect Immun. 2000]

Biosynthesis of ganglioside mimics in *Campylobacter jejuni* OH4384. Identification of the glycosyltransferase genes, enzymatic synthesis of model compounds, and characterization of nanomole amounts by 600-mhz (1)h and (13)c NMR analysis. [J Biol Chem. 2000]

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